

Research Methods:

Site selection

Locations for large-scale restoration activity were determined using a Geographic Information System (GIS) based SAV restoration targeting model (Parham and Karrh 1998). The model uses six layers of key habitat information to evaluate the suitability, ability and potential of a particular habitat to support SAV populations. The data layers incorporated into the targeting model include:

1. **Shoreline:** The Maryland shoreline datalayer used was digitized by the Soil Conservation District using United States Geological Survey (USGS) quad sheets at a scale of 1 inch = 24,000 feet.
2. **Water Quality:** The water quality parameter allows site evaluation based on three methods: Percent light at leaf, percent light at water (Kemp et al., 1995), or the individual water quality parameters (Dennison, 1993). Six water quality parameters important to SAV communities were incorporated into the SAV Restoration Targeting System (light extinction coefficient (Kd), dissolved inorganic nitrogen (DIN), dissolved inorganic phosphorous (DIP), total suspended solids (TSS), chlorophyll a (Chla) and salinity). Data from a running three year growing season (April to October) for SAV were used to obtain a median value by station for each parameter. The data were obtained from the Chesapeake Bay Mainstem and Tributary Water Quality Monitoring Program, intensive surveys, and water quality mapping. The individual water quality parameters were interpolated using the Inverse Distance Weighted interpolation method in Spatial Analyst for ArcView using four nearest neighbors and 100 foot interpolated cells extending beyond the extent of the Chesapeake Bay. After interpolation of the individual parameters, each parameter was overlaid with salinity coverage and assigned as pass or fail based on the SAV habitat requirements for one meter restoration (Batuik et al., 1992).
3. **Bathymetry:** One and two-meter bathymetry contours for the Chesapeake Bay were obtained from the Environmental Protection Agency's, Chesapeake Bay Program, intersected with the Soil Conservation District shoreline and converted from lines to polygons. The resulting shapes were designated to yield areas less than 1 meter depth at mean low water, areas 1 to 2 meters depth and areas greater than 2 meters depth.
4. **Submerged Aquatic Vegetation:** SAV distribution coverage data was determined based on aerial surveys completed by the Virginia Institute of Marine Science (1981-2004). Current distribution was composed of the 2003-2004 SAV distribution. A composite layer of historical SAV distribution was created by combining the 1981, 1984-1990, and 1991-2004 SAV aerial surveys.

5. Hydraulic Clam Dredging: Prohibited clamming areas were mapped based on the laws in the Code of Maryland regulating this activity (§4-1037 and §4-1038). DNR natural oyster bar habitats were buffered by 150 feet as called for in the State and County laws, and a shoreline setback was established and buffered to the appropriate distance (distance varying by County) using the Soil Conservation District Shoreline coverage.

Study area

Five sites in the lower Potomac River were identified as suitable for eelgrass recolonization based on the DNR SAV targeting model (Fig. 3).

- Cherryfield Point (N38° 07.819' W76° 27.574')
- Piney Point (N38° 08.279' W76° 30.159')
- Sage Point (N38° 07 53.2' W076° 26 10.5')
- St. George Island (N38° 08 07.6' W076° 29 41.4')
- Kitt's Point (N36° 06.628' W76° 25.471')

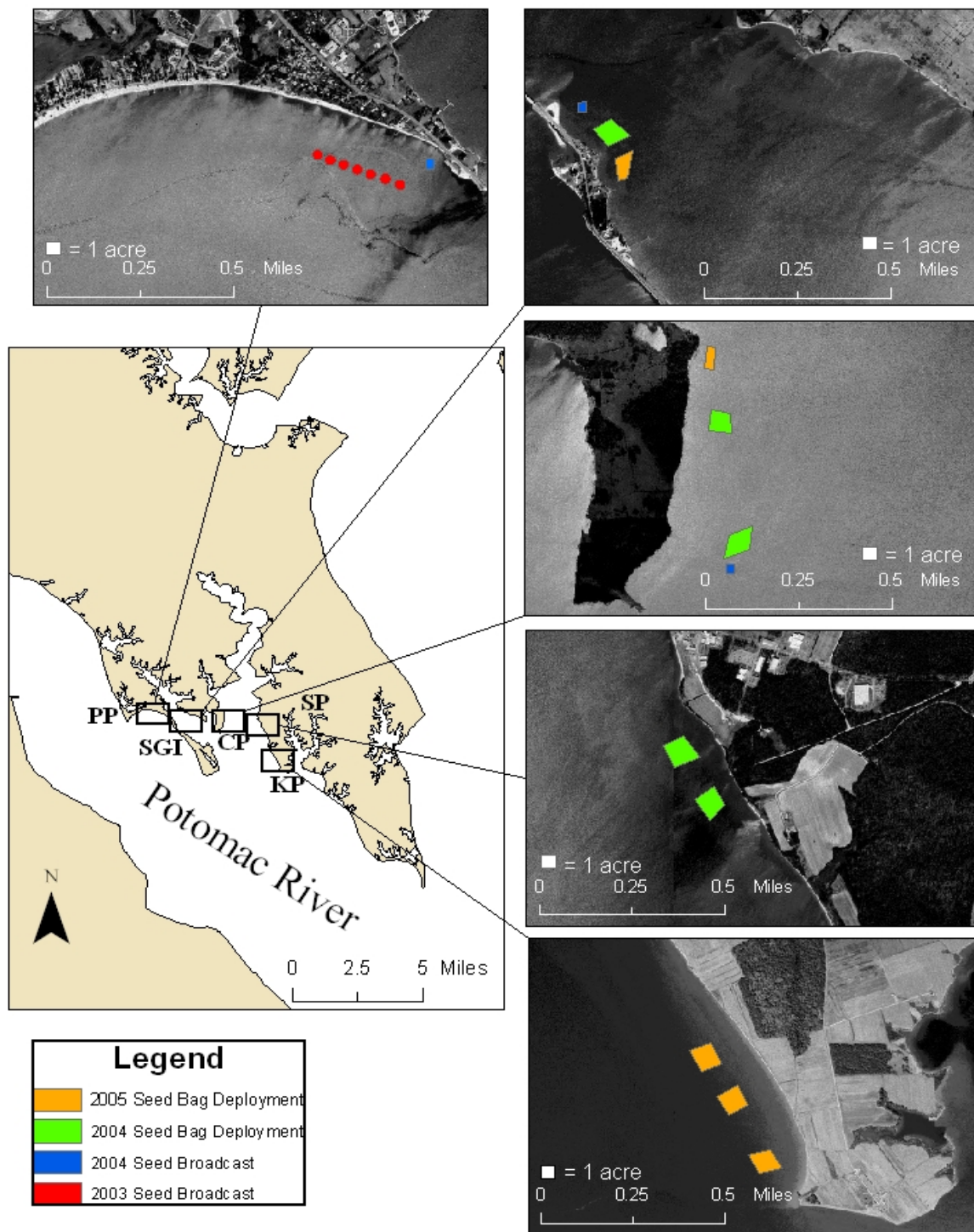


Figure 3. Study area with restoration sites. Cherryfield Point (CP), Piney Point (PP), Sage Point (SP), St. George Island (SGI), and Kitt's Point (KP).

Test plantings

To determine the best planting sites within the areas identified by the SAV restoration targeting model, adults plants raised in the laboratory and harvested from existing beds in the bay were transplanted into three, one square meter plots in areas adjacent to seed broadcast and seed bag areas. Sixty-four adult plants were planted in each plot, anchored by wooden skewers (Davis, 1997). These test plantings were monitored for percent survival at 1 week, 4 weeks and 16 weeks after initial planting.

Adult shoot plantings

As part of the Compensatory Mitigation Package for the Woodrow Wilson Bridge, 20 acres of SAV are being planting by Rummel, Klepper and Kahl (RK&K). The first year's planting of this three year project was 3.5 acres planted in the fall of 2003 with 16,816 planting units (PUs) of widgeon grass (*Ruppia maritima*) and 2,016 PUs of sago pondweed at Sage Point, and 2 acres planted with 15,000 PUs of eelgrass, 964 PUs of sago pondweed and 1,600 PUs of widgeon grass at Piney Point. In spring of 2004, 39,456 sago pondweed PUs were planted at Piney Point. In the fall of 2004 an additional 19,440 PUs of eelgrass, 7,488 PUs of sago pondweed, and 21,479 PUs of widgeon grass were planted at Piney Point, and 2,016 PUs of sago pondweed and 16,816 PUs of widgeon grass were planted at Sage Point.

Seed collection, processing, and storage

Seed Collection

To begin the project, DNR staff concentrated efforts on finding the most productive donor beds from which to harvest. Because the importance of temperature on the life cycle of eelgrass, especially on reproduction, latitudinal



comparisons should show a progression of stages in the reproductive cycle (anthesis and seed release) as one moves south (Silberhorn, 1983). In the Chesapeake Bay's eelgrass beds, anthesis (the period during which a flower is fully open and functional) was observed when temperatures were nearly 15⁰ C, and above 20⁰ C, flowers and immature fruits die and slough off the plant (Silberhorn, 1983).

Figure 4. The mechanical harvester collects eelgrass reproductive shoots.

Since the Chesapeake Bay is close to the southernmost reach of eelgrass distribution, eelgrass flowering and seed release begins in April and May. In 2003, eelgrass reproductive shoots were collected manually from donor beds in Sinepuxent Bay and Tangier Sound. Seed broadcast techniques vary in success with around 15% of viable seeds becoming established (Orth et al., personal communication; Orth et al., 1994), so it is necessary to harvest large numbers of seeds to achieve restoration potential. For approximately 3 weeks, DNR staff and volunteers snorkeled and used scuba equipment to manually remove the reproductive shoots of eelgrass.

This was a very expensive process in terms of man-hours involved, so over the winter, alternative methods of harvesting were investigated. In the past, DNR has contracted the use of a mechanical harvesting boat used for clearing boating channels to harvest water chestnut (*Trapa natans*). It was found that very little work had to be done to adapt this harvester to collect eelgrass reproductive shoots. The reproductive shoots stand above a majority of the plant biomass and could be harvested with little or no impact on the eelgrass beds. During subsequent harvests (2004 and 2005), a mechanical harvest boat was utilized (M J

McCook & Associates, La Plata MD) to increase the volume of reproductive material collected.

Historically, Tangier Sound and the Little Annemessex River have healthy eelgrass beds and in 2004 and 2005 served as donor beds. During aerial surveys of these areas, the areas with the highest density of plants were identified as areas to focus the harvesting efforts. DNR visited each of these donor beds weekly beginning the second week in April. Reproductive structures begin to form when water temperatures reach 10° - 15° C (Granger, 2002). Flowering is completed and viable seeds begin to develop when water temperatures reach 15° -20° C (Granger, 2002). Random samples of reproductive shoots were collected and analyzed to determine the maturity of the seeds.

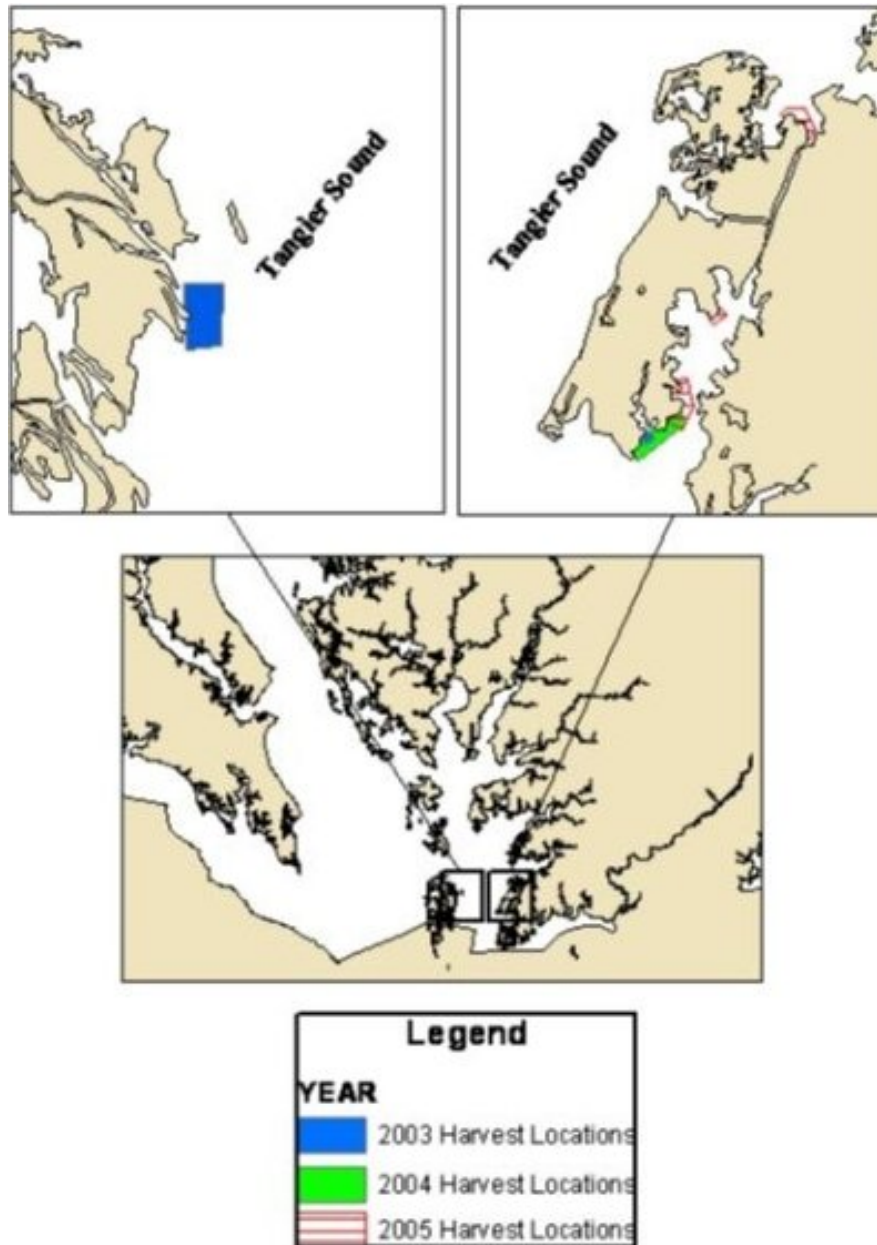


Figure 5. Harvest sites for 2003-2005, Tangier Sound, MD.

When more than 50% of the seeds were mature, and the spathes had begun to drop the seeds, DNR mobilized its field staff to the Tangier Sound eelgrass beds to begin harvesting.

Seeds were collected from donor beds in the Little Annemessex River and Tangier Sound near Smith Island (N37° 58.479' W75° 52.255' and N37° 59.073' W75° 59.206', respectively) and in 2005 from the Little Annemessex River and the mouth of Acre Creek (Big Annemessex River (N37° 59.626' W75° 51.636' and N38° 01.718' W75° 50.632', respectively). The harvester would run systematic transects within the beds, adjusting the cutting blades to account for changes in depth. As the boat moved slowly through the water, the cutting blades clipped the eelgrass reproductive shoots at approximately one foot above the sediment (Fig. 5). The cuttings were sent to the conveyor belt and stored in



Figure 6. Harvesting boat unloading eelgrass.

the hoppers on the back of the boat. Once the boats hoppers were full, a DNR boat would dock with the harvester, and the harvester pilot would unload the cuttings into the DNR boat using a second conveyor belt (Fig. 6).

Once the eelgrass cuttings were on board, biologists and volunteers sorted the material into mesh bags (Fig. 7). The bags were loaded onto a second vessel, which transported the filled bags back to Crisfield, MD. The bags were attached to lines at the dock and kept submerged in the ambient water overnight at Somers Cove marina. Each morning, the bags of harvested material were transported via commercial waterman to the DNR Piney Point Aquaculture facility in St. Mary's County, MD 24-48 hours after collection.



Figure 7 (above). DNR staff and volunteers load the seed material into bags.



Figure 8 (right). Bags of harvested material are loaded onto the transport boat bound for Piney Point.

Surveys done by DNR and VIMS after two years of harvesting concluded that the harvester did not have any significant impacts on the donor beds. Biologists swam through the areas harvested 2 weeks after harvesting, and could not distinguish between harvested and unharvested areas. Aerial surveys in 2005 over the areas harvested in 2004 clearly showed the cutting paths, but no decrease in acreage or plant density was visible.

Seed Processing

Once the bags of harvested eelgrass reproductive shoots arrived at Piney Point Aquaculture Facility, they were placed in one of eight, 20,000 gallon (32'x32'x4') or one of sixteen 9,800 gallon (20'x20'x4') greenhouse basins. The water in each basin was replaced daily with local St. Georges Creek water augmented with aquaculture grade sea salt to match conditions at the harvesting areas (~14ppt). In addition, each basin was aerated to prevent anoxia. Typical basin dissolved oxygen levels averaged 5-6 mg/l. Water quality was monitored twice daily in



Figure 9. Seed settling trays at Piney Point.

order to ensure adequate conditions. While in the basins, the eelgrass seeds slowly dropped from the reproductive shoots over the following month. After all the seeds were released and settled to the bottom of the basins, the seed/reproductive shoot slurry was pumped into a series of stacked settling trays to allow the passive accumulation of seeds while discarding the non-seed material.

Seed Storage

After the completion of seed processing, all seeds were placed in flow-through, aerated and salinity boosted holding tanks until fall seed dispersal. However, slight storage modifications were made each year in order to increase viable seed numbers. In 2004, the seed were stored in a series of three, 2000L cone shaped tanks. Because of the large volume of seeds and the concern for anoxic conditions in poorly mixed seeds, all tanks were heavily aerated to a “rolling boil.” Eelgrass seed storage literature is extremely limited and the group consensus from DNR and VIMS was to try the higher levels of aeration. Extremely low numbers of viable seeds remaining by fall 2004 (~7%) required us to rethink storing seeds in the highly oxygenated system. In 2005, to more closely mimic successful storage conditions developed by VIMS, all seeds were held in a series of ten, 80L shallow tubs with lower aeration and frequent hand mixing to prevent accumulation of silt and possible anoxic conditions. Subsequently, a higher percentage of seeds remained viable for fall dispersal (~25%). While this was an improvement, levels were still below those regularly achieved at VIMS (>50%).

In order to address the issue of low numbers of seeds/ low seed viability, in 2005, a series of seed storage experiments were set up at Piney Point, VIMS (funded by Army Corps of Engineers) and St. Mary’s College to determine optimum storage conditions. The following conditions were examined:

- Source of water
 - River water (filtered, unfiltered),
 - Re-circulated water (filtered, temperature controlled)
- Aeration level – High, low or no air
- Mixing – Seed mixing or no seed mixing
- Bleaching – Bleaching or no bleaching

Based on preliminary results, it appears that the most effective seed storage conditions involve using re-circulated water, with low aeration no mixing. However, we are in the process of evaluating all monitoring data and experimental seed storage methods to determine cause of the poor seed survivability so changes can be made to greatly improve seed viability in 2006.

Seeding Techniques

Seed Bags

A buoy-deployed seeding system (BuDSS) developed by Pickerell et al. was modified slightly and used as an alternative method to broadcasting bare seeds in the fall. There are several potential advantages to using this method, mainly pertaining to not needing to store seeds during the summer. For this method, reproductive material is placed in mesh bags immediately after harvest, moved to the restoration location, and deployed in the area to be restored. Immediate deployment of reproductive material eliminates the need to store seeds, reducing the number of seeds lost to processing and decreasing the expense and labor requirements associated with seed transport, processing, and storage.



As the bags of harvested material arrived at Piney Point, about 15,000 L were used to fill seed bags for deployment. DNR used a modified version of the buoy deployed seeding system, (BuDSS), created by Chris Pickerell at Cornell University Extension Service (Pickerell et al., 2003). Four gallons of collected reproductive shoots were placed in a mesh bag, divided into three sections by cable ties, and supported on each end with a small buoy. At one end, 2.1 m of polypropylene rope was attached to a cinderblock to anchor the seed bag (Fig. 10). The mesh bags suspended above the sediment allowing the seeds to mature and drop over a period of weeks, mimicking natural seeding events (Fig.11). Two types of seed bags were constructed and deployed: single (50,000 seeds) and double (100,000 seeds). Seed bags were deployed at the restoration sites by watermen and DNR staff for approximately one month (Fig.12).

The mesh bags remain suspended at the top of the water column, allowing the seeds to develop and drop over a period of weeks. This mimics the floating and rafting of reproductive shoots during natural seeding events during the natural phenological schedule (Pickerell et al. 2003). Although not proven, it has been suggested that this method may also reduce predation by spreading out seed dispersal over time and through a combination of time and natural forces yield a more even distribution of seeds.

There are potential problems with this method too. These include a navigational hazard while the mesh bags are on-site (restoration plots with floats every 10 meters are difficult to navigate). Despite staggering seed dispersal over time,

seed predators are active during this time. Any sort of spring dispersal that mimics the natural dispersal will be affected by predators.



Figure 10. Picture of seed bag.

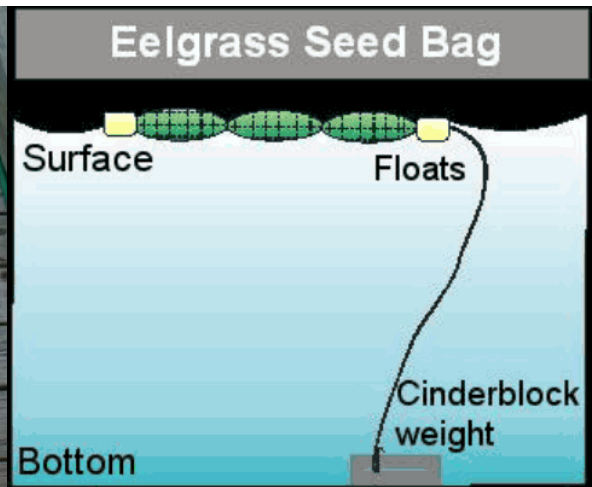


Figure 11. Schematic of seed bag in water column.



Figure 12. Seed bag deployment.

Seed Broadcast

This technique is effective because eelgrass seeds are rapidly incorporated into the sediment and generally do not move far for where they settle (Orth et al 1994). The complexity of the bottom due to biological and physical processes appears to be important to seed retention (Luckenbach and Orth, 1999). Eelgrass

seed recruitment as a percentage of total seeds appears to always be quite low. Annual seed production ranges from 6,176 seed m⁻² to 24,460 seed m⁻² (Olsen 1999) however, even during natural seeding, reported seedling numbers are significantly less than the numbers of seeds produced, ranging from 5-15 percent (Olsen and Sand- Jensen, 1994, Orth, 2003, Granger, 2002, Cook, 1979, Cabin et al., 2000). Researchers using seeds in experimental plantings have encountered varied success, but a common thread seems to be low germination rates (Moore et al, 1993), wash-out of seeds (Orth et al. 1994, Harwell and Orth 1999), and predation (Fishman and Orth, 1996).

Germination in the Chesapeake Bay is thought to be dependent upon temperature, burial, and oxygen cues (Orth and Moore, 1983, Moore 1993). Incorporation of seeds into the sediments (Orth and Moore, 1983, Moore 1993) is essential for the initial of germination. Microtopography prevents long distance redistribution of seeds (Orth et al 1994, Luckenbach and Orth, 1999). Orth et. al. (1994) demonstrated that turbation of the sediment as little as 1 millimeter deep could stop an eelgrass seed from rolling and being transported away. However, deep burial can stop seed germination. Deep burial of seeds below the redox potential discontinuity prevents the developing plant from receiving light (Bigeley, 1981), which may be crucial to germination.

Although not made before the seedings took place, observations by divers during the 2005 surveys at each of the sites suggest that the bottom at each site on the Potomac was suitable for seed recruitment. Seed predation also appears to be an important factor in seed loss (Janzen, 1971, Wassenberg, 1990 and Fishman and Orth, 1996). Experiments where predation was eliminated yielded 100% germination rates illustrating the importance of seed predation (Fishman and Orth, 1996). One mechanism employed by plants to escape predation is to produce seed abundances high enough to satiate the seed predator (Orth, 2003).

Due to the physical presence of three-dimensional structure provided by SAV, and the increased “roughness” of the bottom in SAV beds, water velocities are reduced as much as 50% reduced within SAV beds (Fonseca et. al 1982,, Benoy and Kalff 1999, Gacia et. al 1999). Furthermore, it has been noted that water velocity reductions are directly proportional (as a power function) to both the height and the growth form of the species that occur in the area (Gacia et. al 1999, Petticrew and Kalff 1992).

Eelgrass seeds were hand broadcast using methods used by Orth (Orth, Personal Communication) during the fall of 2003. The restoration site was divided into seven 25 m radius plots 1963.4 m², or 0.485 acres. The plots were divided into 5m concentric circles from a central point. The concentric areas at 5m increments were chosen to evenly allocate the seeds across the plot by

broadcasting while walking around the plot in concentric circles (Fig. 13). To distribute at a density of 100,000 seeds/acre, 50,000 seeds or 660 ml, were broadcast with the appropriate proportions going to each concentric section. For example, 237mL of seeds (36% of the total 660mL) went into the outer ring (green).

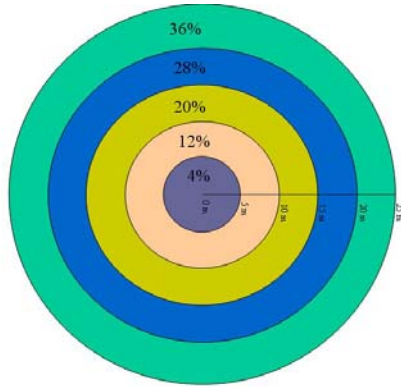


Figure 13. Diagram of the methodology used to disperse seeds by hand using concentric rings (percentages indicate portion of the total seeds distributed in each ring).

This method was slow and did not guarantee an even distribution of seeds. Subsequent seed broadcasts in the fall 2004 were achieved mechanically using a specially designed seed broadcast apparatus developed by C & K Lord and Associates and DNR staff. All seed broadcasts took place before ambient water temperatures dropped to 15°C, the temperature at which eelgrass seeds begin germination. In Maryland, seeds were mechanically dispersed using a newly developed seed-sprayer from C& K Lord, Inc capable of evenly dispersing seeds at suitable densities (200,000 seeds/acre) at the rate of 10 minutes/acre (Fig. 14).

Restoration efforts with eelgrass in plots of different sizes (4 m² to 400m²) and configurations (alternating 4 m² patches and large continuous patches) in different river systems in Virginia (VIMS) have shown a significant site but no significant plot size effect (Orth, personal communication). To look at seeding density effects, Orth et al. (2003) tested five seeding densities ranging from approximately 10,000 seeds/acre to 5,000,000 seeds/acre and found no density dependent effects on germination rate or seedling success. The effectiveness of seeding density was also tested in this project in order to evaluate the potential for site-specific variation in density dependence. Nominal seeding density treatments of 10,000, 50,000, 100,000, and 500,000 seeds/acre were tested. Treatment densities were assigned to randomly chosen plots within the restoration site. The number of treatments, replicates per treatment, and size of plots was dependent upon the number of seeds available. The number of treatments and plot size was reduced as necessary in order to maintain sufficient replication for statistical rigor. After spring surveys (May 2006) the effectiveness

of seeding density will be closely examined to evaluate the potential for site-specific variation in density dependence.



Figure 14. Seed sprayer

Eelgrass spring seed bag and fall seeding sites were located adjacent to State Highway Administration (SHA) sites planted with adult plants at the Piney Point site. Seeds were hand broadcast during the fall of 2003, and by boat in 2004, and 2005. Seeds were broadcast before ambient water temperatures dropped to 15°C, the temperature at which eelgrass seeds begin germination. In Virginia, Orth et al. (2003) tested five seeding densities ranging from approximately 10,000 seeds/acre to 5,000,000 seeds/acre and found no density dependent effects on germination rate or seedling success. The effectiveness of seeding density was also tested in this project in order to evaluate the potential for site specific variation in density dependence. Nominal seeding density treatments of 10,000, 50,000, 100,000, and 500,000 seeds/acre were tested. Treatment densities were assigned to randomly chosen plots within the restoration site. The number of treatments, replicates per treatment and size of plots was dependent upon the number of seeds available. The number of treatments and plot size was reduced as necessary in order to maintain sufficient replication for statistical rigor.

Water Quality Monitoring

The Strategy calls not only for large-scale SAV restoration projects, but also for coincident assessment of the associated habitat conditions in order to evaluate reasons for success or failure and, in turn, improve the likelihood of success of

future projects. DNR conducts temporally and spatially intensive monitoring in Maryland's tidal waters to fully characterize ambient water quality conditions in open and shallow waters. These data have been employed to assess EPA water quality criteria such as dissolved oxygen, chlorophyll, and water clarity, as well as characterize habitat conditions for bay grasses and aquatic organisms. DNR, in association with the Chesapeake Bay Program, has developed consistent monitoring and analysis protocols for these monitoring programs.

Continuous Monitoring

Each continuous monitoring station is equipped with a YSI 6600 water quality monitoring sonde. Beginning in 2004, all YSI 6600 data sondes are equipped with Extended Deployment Systems (EDS). The EDS has a wiper system that allows the continuous monitoring sondes to be deployed for longer periods of time without suffering a degradation of data quality as a result of biofouling. Each continuous monitoring sonde records nine water quality parameters every 15 minutes. The nine water quality parameters measured continuously are water temperature, specific conductance, salinity, dissolved oxygen, turbidity (NTU), fluorescence and total chlorophyll (used to estimate chlorophyll *a*), pH and water depth.

Continuous monitoring sondes are positioned in the water column in either a floating configuration that suspends the sonde at some distance below the surface (usually 1 meter), or in an anchored configuration that fixes the sonde at some distance above the bottom. The sonde position is determined based on the geographic area being monitored and the monitoring goals for that segment. Continuous monitoring sondes in a floating configuration are suspended from a float inside of a 4-inch diameter PVC pipe with 2-inch holes drilled every 4 inches below the waterline to allow for water exchange. Sondes in a fixed configuration are also housed inside a perforated 4-inch diameter PVC pipe, and a bolt is used to hold the negatively buoyant sonde at a fixed depth above the sediment bottom.

Several times a day, the computer server located at the Bay Program office contacts the data logger located at each sampling site via TCP/IP communications and then uploads, archives, and updates the data display on the Eyes on the Bay web site. These data are available immediately on the Internet, allowing the general public to view near real-time water quality data. Details of the steps for installing, calibrating, deploying, and retrieving the YSI instruments are fully provided in DNR's Quality Assurance Project Plan.

In addition to the parameters measured by the sonde, Secchi depth and light attenuation are measured weekly from April to October, and grab samples are taken and filtered on-site or immediately after returning to the laboratory. The

processed samples are sent to the Nutrient Analytical Services Laboratory (NASL) at the Chesapeake Biological Laboratory and to the Maryland Department of Health and Mental Hygiene (DHMH) for analysis. These results are used to analyze relationships between the water quality parameters measured by each continuous monitor and the nutrient component. Some of the lab data were also used to check the YSI data for accuracy. Parameters analyzed at NASL are total dissolved nitrogen, particulate nitrogen, nitrite, nitrite + nitrate, ammonium, total dissolved phosphorus, particulate phosphorus, orthophosphate, dissolved organic carbon, particulate carbon, silicic acid, total suspended solids, volatile suspended solids, particulate inorganic phosphorus and dissolved organic carbon. Parameters analyzed at DHMH include chlorophyll *a*, pheophytin and turbidity.

Water Quality Mapping

Water quality mapping is conducted using water quality mapping, a shipboard system of geospatial equipment and water quality probes that measure water quality parameters from a flow-through stream of water collected near the water's surface. Water quality mapping measures are water temperature, salinity, conductivity, dissolved oxygen, turbidity (NTU), fluorescence (used to estimate chlorophyll *a*) and pH. The water is pumped through a ram (pipe), through the sensors, and then discharged overboard. The water quality mapping unit includes a hand-held Garmin global positioning system (GPS), a microcomputer, and a YSI 6600 sonde with a flow-through chamber. Each water quality datum collected is associated with a date, time, water depth, and GPS coordinate (WGS84).

water quality mapping allows data to be collected rapidly (approximately every four seconds) while the boat is traveling at speeds of up to 25 knots. The water quality mapping system is compact and can fit onto a small boat, allowing sampling in shallow water and the ability to map an entire small tributary such as the Severn River in less than a day. The distance between samples depends on vessel speed; generally at least one observation is collected approximately every 30 meters (100 feet). The water quality mapping system samples water at approximate 0.5-m depths below the surface. Real-time data are displayed in the field through custom software, either in numerical and graphical form or in a real-time mapping application, DATAVIEW, developed by MD DNR.

At 5 to 8 calibration stations per tributary segment, grab samples are collected at 0.5-m depth and filtered on site. The processed samples are sent to the Chesapeake Biological Laboratory and to the DHMH for analysis. Parameters analyzed at NASL total dissolved nitrogen, particulate nitrogen, nitrite, nitrite + nitrate, ammonium, total dissolved phosphorus, particulate phosphorus, orthophosphate, dissolved organic carbon, particulate carbon, silicic acid, total

suspended solids, volatile suspended solids, particulate inorganic phosphorus and dissolved organic carbon.

Parameters analyzed at DHMH include chlorophyll *a*, pheophytin and turbidity. In addition, Secchi depth and photosynthetically active radiation (PAR) measurements are taken at calibration stations to calculate light attenuation (K_d). The calibration station locations are selected to: 1) sample the greatest possible range of water quality conditions found during each cruise; 2) sample a broad spatial area; 3) overlap with long-term fixed monitoring and continuous monitoring stations.

Monitoring Seedling and Vegetative Shoot Success

Surveying and Monitoring

Germination rates, seedling survival, and growth in each seeding density replicate were assessed annually at approximately 1 month, 6 months and 12 months after seeding following methods similar to that of Orth et al. (2003). However, since DNR seeded larger areas than described by Orth et al. (2003), seedling density of seed plots were subsampled by counting the total number of seedlings along diagonal transects between the four corners of the planting area. The areas outside of the original plots were also surveyed to make sure that the broadcast seeds remained within the plots. The number of plants was estimated visually using methods similar to that of Orth et al. (1999). Finally, to determine whether the created eelgrass beds are expanding through vegetative propagation and/or natural seeding, the seed plots and surrounding area were surveyed in the spring and fall following each seeding using aerial overflights and groundtruthing with a handheld mapping GPS.

Test plantings were carried out to ensure that areas identified by the site selection model would support growth of eelgrass. Adult eelgrass plants were transplanted into 3 - 1 m² test plots located adjacent to seed broadcast or seed bag areas. A density of 64 adult plants per m² was used for each test plot. These test plots were monitored at the same time and frequency as the seed plots.